Virtual Entity Yields Real-Time Results

Genomic technologies have revolutionized cancer research, but not without analytical challenges. No single researcher can manage the vast datasets generated by next-generation sequencing and other modern genomic tools. Harnessing those tools in the pursuit of translational advances increasingly compels team science and multidisciplinary collaboration. Toward that end, NCI's Center of Excellence in Integrative Cancer Biology and Genomics (CEICBG) was created in 2008 to unite experts in cancer biology with colleagues in bioinformatics. Working collaboratively, active participants drawn from across NCI use genomics technology to advance basic science discoveries and clinical research applications for the prevention, diagnosis, and treatment of cancer.

Sponsored by CCR, CEICBG is a virtual entity. It exists through the interactions of its members, who are organized into five focus areas (called Subcommittees): Biomarkers and Molecular Targets, Genomics Approaches, Human Genomics and Genetics, Cancer Inflammation, and Integrative/Systems Biology and Bioinformatics. Participating scientists meet regularly to identify potential collaborations within and outside the CEICBG. The group hosts an annual one-day meeting offering enriching lectures and it also hosts a biennial symposium on translational research. The first symposium covered translational genomics, while the second, convened last year, presented clinical applications for next-generation sequencing. It drew more than 1,000 attendees from around the world. The Center plans to host their next symposium in 2014.

"In CEICBG, we investigate cancer in the broadest sense," said Snorri Thorgeirsson, M.D., Ph.D., Chief of CCR's Laboratory of Experimental Carcinogenesis, and Head of the CEICBG, "from tumor biology, to gene expression, to whole genome sequencing, all the way to potential therapies."

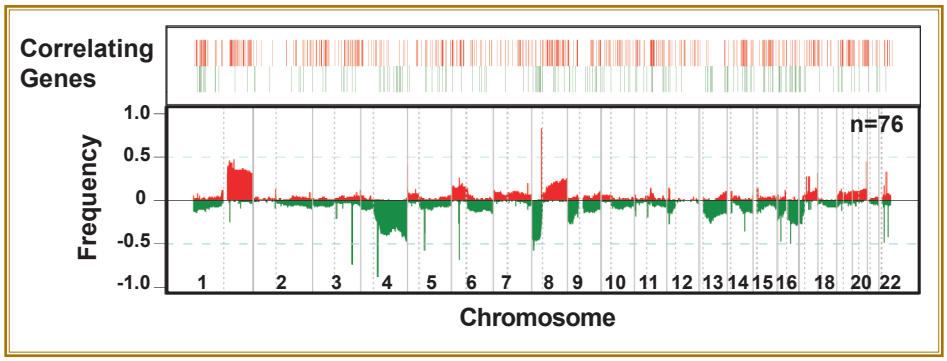
(Photo: R. Baer)

Snorri Thorgerisson, M.D., Ph.D., and Patricia Johnson in CCR's DNA Sequencing Core

Collaborations Enable Insights

CEICBG was deliberately structured to unite scientists with complementary research focus areas. Xin Wang, Ph.D., Deputy Chief of CCR's Laboratory of Human Carcinogenesis (LHC), and Thomas Ried, M.D., a Senior Investigator in CCR's Genetics Branch, both members of the Biomarkers and Molecular Targets Subcommittee, collaborate on research projects to develop and validate biomarkers in cancer treatment that offer an

(Image: X. Wang, Ph.D.)



In the somatic cells from 76 clinical specimens, significant differences in gene copy numbers were noted at specific locations in the genome. Frequencies of samples showing copy number increases at a particular site are shown in red, and those showing copy number decreases are shown in green.

excellent example of CEICBG's fruitfulness.

Wang explains how he and Ried study genomic data—which is generated in his laboratory—to segregate tumors into discreet clusters of biomarkers that may eventually help clinicians match the right drugs with the right patients. Biomarker research may one day differentiate "driver" mutations that promote tumor growth from "passenger mutations" that do not. "If we can identify druggable tumor drivers, we can eliminate tumors completely," Wang said.

For help with his data analysis, Wang turns to Paul Meltzer, M.D., Ph.D., Chief of CCR's Genetics Branch and a member of the Genomics Approaches Subcommittee. Throughout the CEICBG, Meltzer shares new analytic technologies with his colleagues. Recently, he and Wang pinpointed driver mutations in liver cancer by looking at changes in a parameter called somatic copy number alteration. As Meltzer explains, normal cells ordinarily have two copies of each gene, one inherited from each parent. But because tumors are genomically unstable, copy numbers can vary in cancer—tumor suppressor genes may be deleted, for example, while genes that drive cancer progression may be amplified, so hundreds of copies may be present.

In their collaboration—published last year in *Gastroenterology*—Wang and Meltzer measured copy number changes to find driver mutations involving 10 genes on chromosome 8p, which predict poor outcomes. They also concluded that the expression pattern of these driver genes could be useful in tumor diagnosis and staging.

Wang's collaborations with CEICBG members and researchers in China have already produced new diagnostics for liver cancer that are now being evaluated in clinical trials; one is in China and the other in the U.S. (clinicaltrials.gov: NCT01681446). The first trial relies on assays that predict the response to interferon-alpha therapy based on the expression of microRNA-26, which is a small, non-coding RNA with tumor-suppressor function. Patients who express low levels of this biomarker respond far better to the drug than those with higher levels, Wang's research has shown. "So this is a good example of precision medicine," he said. The trial in the U.S. screens for a specific gene signature that predicts tumor relapse. Described in a 2010 paper in *Cancer Research*, grouping of 161 genes predicts overall and disease-free survival, which is especially useful for patients with early-stage disease. The diagnostic incorporates those findings, Wang said, and is used to select patients for additional and aggressive adjuvant therapy.

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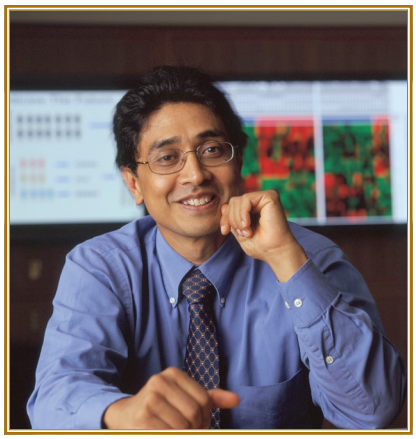
The Human Genomics and Genetics focus area fosters collaborations aimed at finding harmful gene variants through genome-wide association studies (GWAS). Curt Harris, M.D., Chief of LHC, says one of his team's ongoing projects reflects the Subcommittee's mission. Together with scientists from across NCI and beyond, Harris and colleagues are conducting the first GWAS study of lung cancer in African Americans. This population has disproportionately higher rates of lung cancer and also has a more aggressive form of the disease.

GWAS studies are often large (the current lung cancer study enrolls roughly 5,000 people), so the team works with collaborators located throughout the U.S. who collect tissue

(Photo: R. Baer)



Javed Khan, M.D.

samples and clinical data. One of their NCI research partners is Stephen Chanock, M.D., in NCI's Division of Cancer Epidemiology and Genetics (DCEG). Chanock's laboratory handles the analytical component of the study—specifically, by looking for genetic variants in lung tissue—while Harris's team handles the integrative molecular epidemiology, which combines molecular genetics with traditional epidemiology. "We are looking for unique variants in African Americans that we can use for diagnostic and prognostic purposes," said Harris. "And that will lead to functional analyses focused on understanding how these variants contribute to tumor risk and progression."

The Integrative/Systems Biology and Bioinformatics focus area applies a range of analytical and bioinformatic approaches to find cancer biomarkers for use in the clinic and in drug development. "We try to integrate high-throughput "omic" data into a coherent story of what is happening in the cancer cell," said Javed Khan, M.D., a Senior Investigator in CCR's Pediatric Oncology Branch (POB). "That tends to involve a lot of high-end computational analysis and integration of large datasets. Ideally, we can identify the genes and pathways that drive a particular cancer, and then that leads to potential treatments."

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Khan's laboratory is now engaged in a new study opening soon that illustrates this approach. His collaborator is Brigitte Widemann, M.D., also a POB Senior Investigator, who works on a rare and aggressive illness known as malignant peripheral nerve sheath tumor (MPNST). This cancer of connective tissues surrounding the nerves often arises in pediatric patients with a germline *NF1* mutation on chromosome 17. All patients with *NF1* mutations develop neurofibromatosis type 1; a condition that produces skin spots, benign and/or cancerous nerve tumors, bone abnormalities, and other symptoms. Roughly 25-40 percent of *NF1* mutant carriers also develop plexiform neurofibromas, which are noncancerous nerve tumors that cause pain and disfigurement. Plexiform neurofibromas can also transition to MPNST, which is often fatal and for which the only available standard treatment is surgery.

Together with Douglas Stewart, M.D., in DCEG, Widemann and Khan hope to find biomarkers that predict the malignant shift from plexiform neurofibroma to MPNST. Widemann's role will be in the clinical evaluation of the patients, and the selection of suspect lesions for biopsy, as well as adjacent tissues that she believes might remain free of the cancer. She will then turn those biopsies, along with blood samples, over to Stewart and Khan. And they, in turn, will sequence the tissues and perform additional bioinformatic analyses aimed at finding predictive biomarkers, and possibly targets for therapy. Widemann's enthusiasm is evident as she describes her vision for the future, "This is optimal sharing of mutually beneficial expertise. We run one of the largest *NF1* clinics in the country, so we can do the expert phenotyping while Khan and Stewart do the expert genomic studies and biological validation."

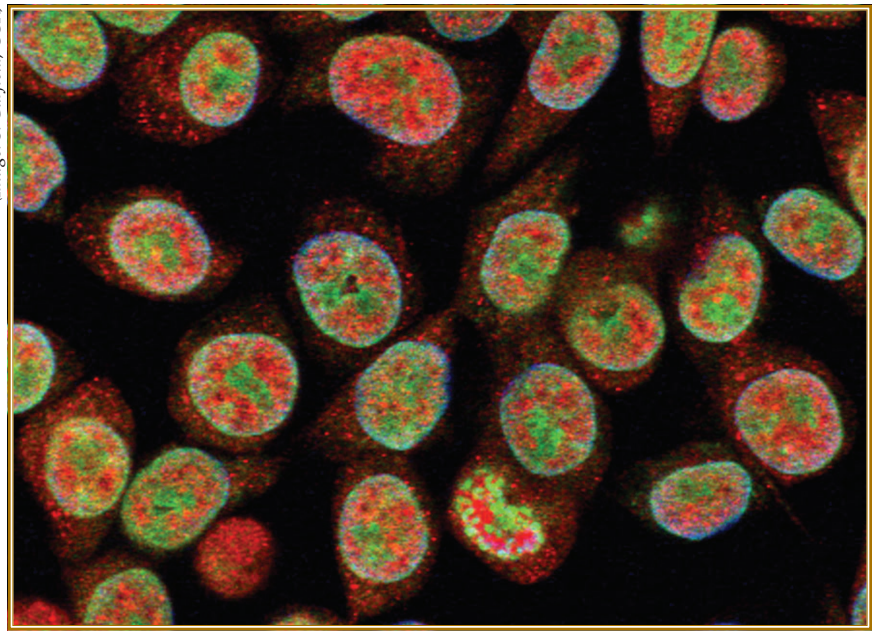
Core Facilities Offer Analytical Support

In addition to fostering collaborations, the CEICBG works at expanding use of the core facilities that NCI researchers rely on for sequencing and analytical support. According to David Goldstein, Ph.D., Head of CCR's Office of Science and Technology Partnerships, the Sequencing Facility run by SAIC, which supplies support and assistance at each phase of the sequencing process, is important in that respect. "This facility has made a significant impact on research at CCR," Goldstein said. The Sequencing Facility offers the latest in massively-parallel technologies, which allow them to generate whole genome analyses in a matter of days.

CCR researchers use the core to study the heterogeneity that exists within and between tumors and to examine many other aspects related to tumor biology: They discover how genomes are rearranged and altered, which regulators are active, and how epigenetic changes affect the cell. And all of this helps them to identify potential drug targets.

But between the sequencing and its therapeutic promise, Goldstein explains, lies a major bottleneck: Enormous and often overwhelming

(Image: S. Garfield, CCR)



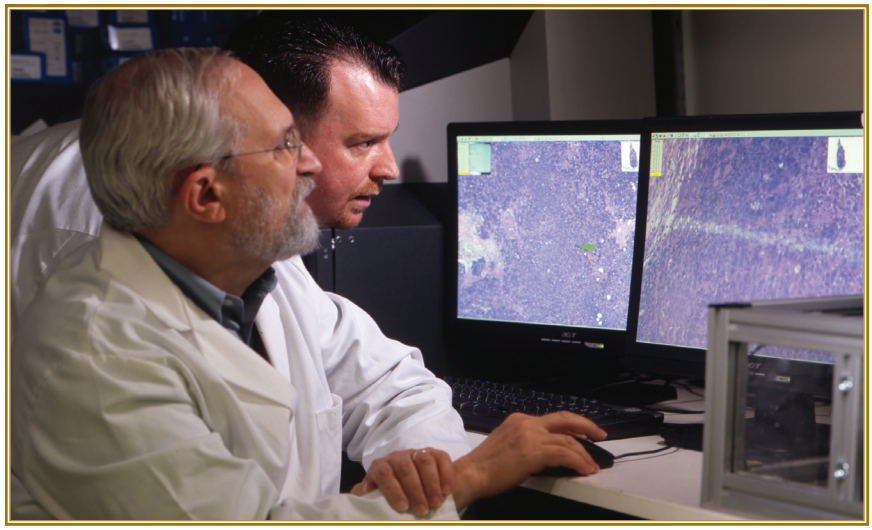
Confocal microscopy image of colon carcinoma.

amounts of data. To address that issue, Goldstein's office helped create the CCR Bioinformatics Core, which was launched in January 2011 to provide analytical support to CCR scientists who do not otherwise have access to bioinformatics. By increasing understanding of bioinformatics techniques and processes among CCR scientists, this Core empowers them to perform basic, informed analyses for their research projects.

Similarly, Meltzer directs the Clinical Molecular Profiling Core, which facilitates the collection of biological data on tumors entered into a CCR clinical trial. Available to scientists throughout NCI, the core offers a range of analytical capabilities: gene expression profiling, comparative genomic hybridization, high-density single-nucleotide polymorphism analysis, and more. The Core spares clinical investigators from having to learn how to use these technologies on their own.

Yet another technology resource, the Confocal Microscopy Core Facility, offers capabilities for tracking live cells and cell components over time—even when in living animals. Confocal microscopy uses lasers to track fluorescently-tagged proteins, cells, and tissues. Available to all NCI staff, the technology's advantage lies in its ability to image specific cells and biomolecules; this is unlike other high-throughput analytical tools that take average measures from cells and tissues mixed together, explains Susan Garfield, M.S., the core's director. Applied to cancer research, scientists use the technology to study protein expression patterns and markers that might change during tumor evolution.

(Photo: R. Baer)



Paul Meltzer, M.D., Ph.D., and colleague

Collaborations Share a Vision

CEICBG's main goal is to use advanced analytic technology to define homogenous clusters of patients, who can then be treated with appropriate therapies. "This is what everyone in the field wants to do," Thorgeirsson said. "So in that sense, CEICBG is a vehicle within CCR and NCI that drives and accelerates translational applications built upon the immense amount of basic research data available here." By bringing expertise in a number of scientific focus areas together to advance cancer research, CEICBG shortens the time between discovery and patient benefit.

For more information about the NCI Center of Excellence in Integrative Cancer Biology and Genomics, please visit its Web site at https://ccrod.cancer.gov/confluence/display/COEICBG/Home.